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PHM. 98-149/GB/P

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3. Full name, address and postcode of the or of each applicant *(underline all surnames)*

ZENECA Limited
15 Stanhope Gate
London W1Y 6LN
GB

Patents ADP number *(if you know it)*

6254007002

If the applicant is a corporate body, give the country/state of its incorporation

4. Title of the invention

CHEMICAL COMPOUNDS

5. Name of your agent *(if you have one)*

DENERLEY, Paul Millington

"Address for service" in the United Kingdom to which all correspondence should be sent *(including the postcode)*

Intellectual Property Department
ZENECA Pharmaceuticals
Mereside, Alderley Park
Macclesfield
Cheshire
SK10 4TG, GB

Patents ADP number *(if you know it)*

1030618002

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Country	Priority application number *(if you know it)*	Date of filing *(day / month / year)*

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Number of earlier application	Date of filing *(day / month / year)*

8. Is a statement of inventorship and of right to grant of a patent required in support of

CHEMICAL COMPOUNDS

The present invention relates to chemical compounds, to their production as well as to pharmaceutical compositions containing them as well as to their use in therapy, in particular of inflammatory disease.

MCP-1 is a member of the chemokine family of pro-inflammatory cytokines which mediate leukocyte chemotaxis and activation. MCP-1 is a C-C chemokine which is one of the most potent and selective T-cell and monocyte chemoattractant and activating agents known. MCP-1 has been implicated in the pathophysiology of a large number of inflammatory diseases including rheumatoid arthritis, glomerular nephritides, lung fibrosis, restenosis (International Patent Application WO 94/09128), alveolitis (Jones et al., 1992, *J. Immunol.*, **149**, 2147) and asthma. Other disease areas where MCP-1 is thought to play a part in their pathology are atherosclerosis (e.g. Koch et al., 1992, *J. Clin. Invest.*, **90**, 772 -779), psoriasis (Deleuran et al., 1996, *J. Dermatological Science*, **13**,. 228-236), delayed-type hypersensitivity reactions of the skin, inflammatory bowel disease (Grimm et al., 1996, *J. Leukocyte Biol.*, **59**,. 804-812), multiple sclerosis and brain trauma (Berman et al, 1996, *J. Immunol.*, **156**,. 3017-3023). An MCP-1 inhibitor may also be useful to treat stroke, reperfusion injury, ischemia, myocardial infarction and transplant rejection.

MCP-1 acts through the MCP-1 receptor (also known as the CCR2 receptor). MCP-2 and MCP-3 may also act, at least in part, through the MCP-1 receptor. Therefore in this specification, when reference is made to "inhibition or antagonism of MCP-1" or "MCP-1 mediated effects" this includes inhibition or antagonism of MCP-2 and/or MCP-3 mediated effects when MCP-2 and/or MCP-3 are acting through the MCP-1 receptor.

Copending International Patent Application Nos. PCT/GB98/02340 and PCT/GB98/02341 describe and claim groups of compounds based upon the indole ring structure which are inhibitors of MCP-1 and therefore have applications in therapy.

The use of certain indole derivatives as NMDA antagonists is described is USP5051442, WO9312780, EP-483881. Other indoles and their use as inhibitors of [illegible] described in for example EP-A-275-667

R^3 is hydrogen, a functional group, optionally substituted alkyl, optionally substituted alkenyl, optionally substituted alkynyl, optionally substituted aryl, optionally substituted heterocyclyl, optionally substituted alkoxy, optionally substituted aralkyl, optionally substituted aralkyloxy, optionally substituted cycloalkyl;

R^4 is a group $NHCOR^{15}$, $NHSO_2R^{15}$ or $OCONR^{16}R^{17}$ where R^{15} is optionally substituted alkyl, optionally substituted aryl or optionally substituted heteroaryl and R^{16} and R^{17} are independently selected from hydrogen, optionally substituted alkyl, optionally substituted aryl and optionally substituted heteroaryl, with the proviso that at least one of R^{16} or R^{17} is other than hydrogen, or R^{16} and R^{17} together with the nitrogen atom to which they are attached form an optionally substituted heterocyclic ring which optionally contains further heteroatoms; and

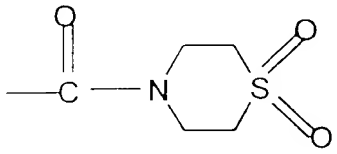
R^5, R^6 and R^7 are independently selected from hydrogen, a functional group or an optionally substituted hydrocarbyl groups or optionally substituted heterocyclic groups.

Compounds of formula (I) are inhibitors of monocyte chemoattractant protein-1. In addition, they appear to inhibit RANTES induced chemotaxis. RANTES is another chemokine from the same family as MCP-1, with a similar biological profile, but acting though the CCR1 receptor. As a result, these compounds can be used to treat disease mediated by these agents, in particular inflammatory disease.

In this specification the term 'alkyl' when used either alone or as a suffix includes straight chained, branched structures. These groups may contain up to 10, preferably up to 6 and more preferably up to 4 carbon atoms. Similarly the terms "alkenyl" and "alkynyl" refer to unsaturated straight or branched structures containing for example from 2 to 10, preferably from 2 to 6 carbon atoms. Cyclic moieties such as cycloalkyl, cycloalkenyl and cycloalkynyl are similar in nature but have at least 3 carbon atoms. Terms such as "alkoxy" comprise alkyl groups as is understood in the art.

The term "halo" includes fluoro, chloro, bromo and iodo. References to aryl groups include aromatic carbocylic groups such as phenyl and naphthyl. The term "heterocyclyl" includes aromatic or non-aromatic rings, for example containing from 4 to 20, suitably from 5 [illegible] heteroatom such as oxygen, sulphur or nitrogen

optionally substituted with alkyl, aryl or aralkyl. A specific functional group which is suitable for R^4, R^5, R^6 and/or R^7 is a group of sub-formula (IV).



(IV)

Particular examples of groups R^5, R^6 and R^7 are hydrogen, hydroxy, halo or alkoxy. In particular R6 and R^7 are hydrogen. R^5 may be hydrogen but in addition is suitably a small subsitutent such as hydroxy, halo or methoxy.

Particular substituents for R^1 include trifluoromethyl, C_{1-4}alkyl, halo, trifluoromethoxy, C_{1-4}alkoxy, C_{1-4}alkanoyl, C_{1-4}alkanoyloxy, nitro, carbamoyl, C_{1-4}alkoxycarbonyl, C_{1-4}alkylsulphanyl, C_{1-4}alkylsulphinyl, C_{1-4}alkylsulphonyl, sulphonamido, carbamoylC_{1-4}alkyl, *N*-(C_{1-4}alkyl)carbamoylC_{1-4}alkyl, *N*-(C_{1-4}alkyl)$_2$carbamoyl-C_{1-4}alkyl, hydroxyC_{1-4}alkyl or C_{1-4}alkoxyC_{1-4}alkyl.

Additionally or alternatively, two such substituents together may form a divalent radical of the formula -O(CH_2)$_{1-4}$O- attached to adjacent carbon atoms on the R^1 ring.

Preferred substitutents for R^1 are one or more non-polar substituents such as halo.

In particular, R^1 is substituted by one or more halo groups, in particular chlorine. A particular example of an R^1 group is 3,4-dichlorophenyl, 3-fluoro-4-chlorophenyl, 3-chloro-4-fluorophenyl or 2,3-dichloropyrid-5-yl.

Examples of groups R^2 include carboxy; cyano; tetrazol-5-yl; SO_3H; -CONHR^8 where R^8 is selected from cyano, hydroxy, -SO_2R^{12} where R^{12} is alkyl such as C_{1-4} alkyl, aryl such as phenyl, heteroaryl or trifluoromethyl, or R^8 is a group-(CHR^{10})$_r$-COOH where r is an integer of 1-3 and each R^{10} group is independently selected from hydrogen or alkyl such as C_{1-4} alkyl; or R^2 is a group -SO_2NHR^9 where R^9 is an optionally substituted phenyl or an optionally substituted 5 or 6 membered heteroaryl group, or a group COR^{14} where R^{14} is alkyl such as

pyridyl; pyrimidinyl; phenyl optionally substituted by halo such as chloro, hydroxy, alkoxy such as methoxy, carbamoyl, acyl such as acetyl, or hydroxyalkyl where the alkyl group suitably includes at least two carbon atoms, such as hydroxyethyl.

Where R^{15}, R^{16} and/or R^{17} is a heterocyclyl group, or where R^{16} and R^{17} together form an optionally substituted heterocyclic ring, these may be substituted by functional groups such as halo or hydroxy, or by alkyl groups such as methyl or ethyl, or alkenyl or alkynyl groups any of which may be subsituted, for example with hydroxy, as well as with further heteroaryl groups such as pyridyl.

Particular examples of R^{15} include alkyl in particular methyl optionally substituted by a functional group or, in particular, a heterocylcyl group where the heterocyclyl group may be optionally substituted by a functional group such as halo or hydroxy or by an alkyl group such as methyl. Other examples of R^{15} are heterocylcyl groups which are optionally subsitituted for example by alkyl such as methyl, functional groups such as chloro or heterocycyl groups such as pyridyl.

Particular examples of R^{16} and R^{17} are alkyl such as methyl.

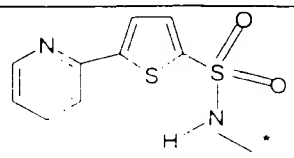
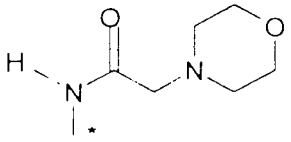
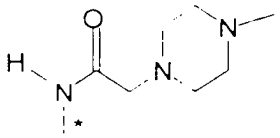
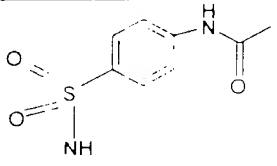
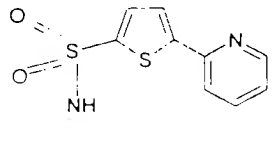
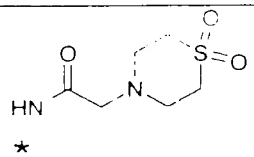
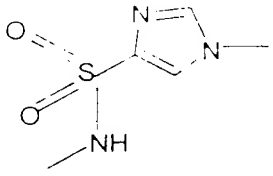
X is CH_2 or SO_2 and is preferably CH_2.

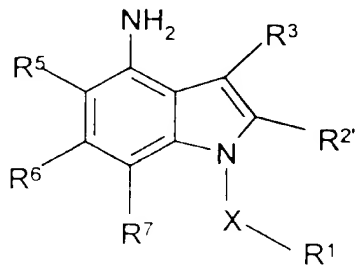
Suitable pharmaceutically acceptable salts of compounds of formula (I) include acid addition salts such as methanesulfonate, fumarate, hydrochloride, hydrobromide, citrate, maleate and salts formed with phosphoric and sulphuric acid. In another aspect suitable salts are base salts such as an alkali metal salt for example sodium, an alkaline earth metal salt for example calcium or magnesium, an organic amine salt for example triethylamine, morpholine, *N*-methylpiperidine, *N*-ethylpiperidine, procaine, dibenzylamine, *N,N*-dibenzylethylamine or amino acids for example lysine. There may be more than one cation or anion depending on the number of charged functions and the valency of the cations or anions. A preferred pharmaceutically acceptable salt is a sodium salt.

An *in vivo* hydrolysable ester of a compound of the formula (I) containing carboxy or hydroxy group is, for example, a pharmaceutically acceptable ester which is hydrolysed in the human or animal body to produce the parent acid or alcohol.

[illegible] suitable for carboxy include alkyl esters such as

esters, C_{3-6}cycloalkoxycarbonyloxyC_{1-6}alkyl esters for example

Compd No.	R^3	R^4	R^5	R^6	R^a	R^b
1	H		H	H	H	H
2	H		H	H	Cl	Cl
3	H		H	H	Cl	Cl
4	H		H	H	Cl	Cl
5	H		H	H	Cl	Cl
6	H		H	H	Cl	Cl
7	H		H	H	Cl	Cl
8	H	$NHC(O)CH_2NHCH_2COOH$	H	H	Cl	Cl



(VII)

where X, R^1, R^3, R^5, R^6 and R^7 are as defined in relation to formula (I), R^2 is a group R^2 as defined in relation to formula (I) or a protected form thereof, with a compound of formula (VIII)

$$Z\text{-}R^{22}$$

(VIII)

where Z is a leaving group and R^{22} is a group $COR^{15'}$ or $SO_2R^{15'}$ where $R^{15'}$ is group R^{15} as defined in relation to formula (I) or a precursor thereof;

and thereafter if desired or necessary:

(i) converting a precursor group $R^{15'}$ to a group R^{15} and/or converting a group R^{15} to a different such group;

(ii) deprotecting a group $R^{2'}$ to a group R^2.

Suitable leaving groups Z include halo such as chloro.

The reaction is suitably effected in an organic solvent such as dichloromethane or tetrahydrofuran in the presence of a base such as triethylamine or pyridine. Moderate temperatures, for example from 0° to 50°C and conveniently ambient temperature, are employed in the reaction.

Compounds of formula (I) where R^4 is a group $OCONR^{16}R^{17}$ may be prepared by a broadly similar method by reacting a compound of formula (VIIA)

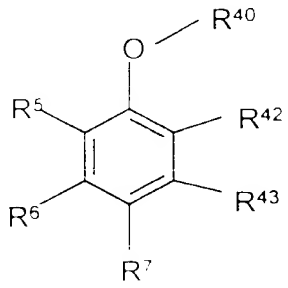
Suitable leaving groups for Z include halide such as chloride, bromide or iodide, as well as mesylate or tosylate. The reaction is suitably effected in an organic solvent such as dimethylformamide (DMF) tetrahydrofuran (THF) or DCM in the presence of a base such as sodium hydride, sodium hydroxide, potassium carbonate. Optionally the reaction is effected in the presence of a suitable phase transfer catalyst. The choice of base and solvent is interdependent to a certain extent in that certain solvents are compatible with some bases only as is understood in the art. For example, sodium hydride may preferably be used with dimethylformamide or tetrahydrofuran and sodium hydroxide is preferably used with dichloromethane and a phase transfer catalyst.

The reaction can be carried out at moderate temperatures, for example from 0 to 50°C and conveniently at about ambient temperature.

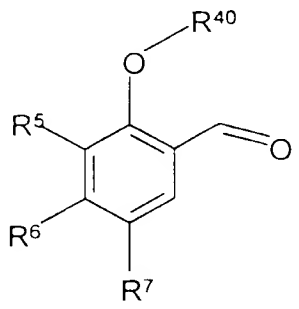
Preferably, $R^{2'}$ is an ester group in the compound of formula IX and this may be subsequently converted to an acid or to another ester or salt, by conventional methods later in the process. For example, when X is a group SO_2 and R^2 is a methyl ester of carboxy, it may be converted to the corresponding carboxylic acid by reaction with lithium iodide in dry pyridine or DMF.

Suitable protecting groups R^{40} include acetyl or benzyl. The reaction conditions employed will be variable depending upon the nature of the protecting group R^{40} and would be apparent to a skilled person. Acetyl groups may be removed by reaction with a strong base such as sodium methoxide, whereas benzyl groups may be removed by hydrogenation for example in the presence of a catalyst such as a palladium catalyst.

Compounds of formula (IX) may be prepared by cyclisation of a compound of formula (XII)



Compounds of formula (XIII) where R^3 is hydrogen may be prepared for example by reacting a compound of formula (XV)



(XV)

with a compound of formula (XVI)

$$N_3CH_2R^{2'}$$

(XVI)

where R^5, R^6, R^7, and $R^{2'}$ are as defined hereinbefore. The reaction may be effected in an organic solvent such as ethanol at low temperatures of from -20 to 0°C, suitably at about 0°C. The reaction is suitably effected in the presence of a base such as an alkoxide, in particular an ethoxide, for example potassium ethoxide.

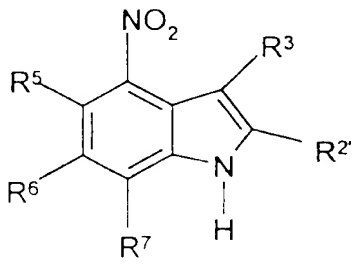
Compounds of formula (XVI) are suitably prepared by reacting a compound of formula (XVII)

$$R_{47}CH_2R^{2'}$$

(XVII)

where $R^{2'}$ is defined above and R^{47} is a leaving group such as halide and in particular bromide, with an azide salt, such as an alkali metal azide salt in particular sodium azide.

Compounds of formula (XIV) may be prepared by reacting a compound of formula (XVIII)



(XX)

where $R^{2'}$, R^3, R^5, R^6 and R^7 are as defined above.

Compounds of formula (X), (XVI), (XVII), (XVIII), (XIX) and (XX) are either known compounds or they may be prepared from known compounds by conventional literature methods.

According to a further aspect of the invention there is provided a compound of the formula **(I)** as defined herein, or a pharmaceutically acceptable salt or an *in vivo* hydrolysable ester thereof, for use in a method of treatment of the human or animal body by therapy. In particular, the compounds are used in methods of treatment of inflammatory disease.

According to a further aspect of the present invention there is provided a method for antagonising an MCP-1 mediated effect in a warm blooded animal, such as man, in need of such treatment, which comprises administering to said animal an effective amount of a compound of formula **(I)**, or a pharmaceutically acceptable salt, or an *in vivo* hydrolysable ester thereof.

The invention also provides a pharmaceutical composition comprising a compound of formula **(I)** as defined herein, or a pharmaceutically acceptable salt, or an *in vivo* hydrolysable ester thereof, in combination with a pharmaceutically acceptable diluent or carrier.

The compositions of the invention may be in a form suitable for oral use (for example as tablets, lozenges, hard or soft capsules, aqueous or oily suspensions, emulsions, dispersible powders or granules, syrups or elixirs), for topical use (for example as creams, ointments, gels, or aqueous or oily solutions or suspensions), for administration by inhalation (for example as a finely divided powder or a liquid aerosol), for administration by insufflation (for example as a finely divided powder) or for parenteral administration (for example as a sterile

The compositions of the invention may be prepared by conventional methods using conventional pharmaceutical excipients, well known in the art. Thus, compositions intended

Oily suspensions may be formulated by suspending the active ingredient in a vegetable oil (such as arachis oil, olive oil, sesame oil or coconut oil) or in a mineral oil (such as liquid paraffin). The oily suspensions may also contain a thickening agent such as beeswax, hard paraffin or cetyl alcohol. Sweetening agents such as those set out above, and flavouring agents may be added to provide a palatable oral preparation. These compositions may be preserved by the addition of an anti-oxidant such as ascorbic acid.

Dispersible powders and granules suitable for preparation of an aqueous suspension by the addition of water generally contain the active ingredient together with a dispersing or wetting agent, suspending agent and one or more preservatives. Suitable dispersing or wetting agents and suspending agents are exemplified by those already mentioned above. Additional excipients such as sweetening, flavouring and colouring agents, may also be present.

The pharmaceutical compositions of the invention may also be in the form of oil-in-water emulsions. The oily phase may be a vegetable oil, such as olive oil or arachis oil, or a mineral oil, such as for example liquid paraffin or a mixture of any of these. Suitable emulsifying agents may be, for example, naturally-occurring gums such as gum acacia or gum tragacanth, naturally-occurring phosphatides such as soya bean, lecithin, an esters or partial esters derived from fatty acids and hexitol anhydrides (for example sorbitan monooleate) and condensation products of the said partial esters with ethylene oxide such as polyoxyethylene sorbitan monooleate. The emulsions may also contain sweetening, flavouring and preservative agents.

Syrups and elixirs may be formulated with sweetening agents such as glycerol, propylene glycol, sorbitol, aspartame or sucrose, and may also contain a demulcent, preservative, flavouring and/or colouring agent.

The pharmaceutical compositions may also be in the form of a sterile injectable aqueous or oily suspension, which may be formulated according to known procedures using one or more of the appropriate dispersing or wetting agents and suspending agents, which have been mentioned above. A sterile injectable preparation may also be a sterile injectable solution or suspension in a non-toxic parenterally-acceptable diluent or solvent, for example a [illegible] in 1,3-butanediol

The size of the dose for therapeutic or prophylactic purposes of a compound of the Formula I will naturally vary according to the nature and severity of the conditions, the age and sex of the animal or patient and the route of administration, according to well known principles of medicine. As mentioned above, compounds of the Formula I are useful in treating diseases or medical conditions which are due alone or in part to the effects of farnesylation of rats.

In using a compound of the Formula I for therapeutic or prophylactic purposes it will generally be administered so that a daily dose in the range, for example, 0.5 mg to 75 mg per kg body weight is received, given if required in divided doses. In general lower doses will be administered when a parenteral route is employed. Thus, for example, for intravenous administration, a dose in the range, for example, 0.5 mg to 30 mg per kg body weight will generally be used. Similarly, for administration by inhalation, a dose in the range, for example, 0.5 mg to 25 mg per kg body weight will be used. Oral administration is however preferred.

A further aspect of the invention comprises the use of a compound of formula (I) as defined above in the preparation of a medicament for the treatment of inflammatory disease.

The invention is further illustrated, but not limited by the following Examples in which the following general procedures were used unless stated otherwise.

Preparation 1

Ethyl *N*-(3,4-dichlorobenzyl)-4-nitroindole-2-carboxylate

Ethyl 4-nitroindole-2-carboxylate (26 g) [prepared according to S. M. Parmerter *et. al.* *J. Amer. Chem. Soc.*, 1958, **80**, 4621], 3,4-dichlorobenzyl chloride (16 ml), potassium carbonate (17 g) and potassium iodide (2 g) in DMF (250 ml) were stirred at 60°C for 2 hours. The reaction was concentrated *in vacuo* and the residue partitioned between water and dichloromethane. *I*so-hexane was added to the combined organic extracts resulting in crystallisation of the product as yellow needles (39 g, 89%) NMR d (CD_3SOCD_3) 1.30 (t, 3H), 4.32 (q, 2H), 5.93 (s, 2H), 6.88 (dd, 1H), 7.18 (d, 1H), 7.52 (d, 1H), 7.56 (dd, 1H), 7.78 (s, [illegible] 2H); MS (AP+) 393

(1.98 g, 89%); NMR d (CD_3SOCD_3) 1.3 (t, 3H), 4.2 (q, 2H), 5.7 (s, 4H), 6.2 (d, 1H), 6.6 (d, 1H), 7.0 (m, 2H), 7.25 (m, 1H),
7.5 (d, 1H), 7.6 (m, 1H): *M*/z (+) 363.3 (MH^+).

Preparation 4

Ethyl 4-chloroacetamido-*N*-(3,4-dichlorobenzyl)indole-2-carboxylate

Ethyl 4-amino-*N*-(3,4-dichlorobenzyl)indole-2-carboxylate (2.03 g), chloroacetyl chloride (0.5 ml) and triethylamine (4.0 ml) were stirred in dichloromethane (50 ml) for 16 hours. The reaction was washed with water, dried ($MgSO_4$) and concentrated *in vacuo*. The residue was triturated with toluene to give the product as a pale grey solid (1.61 g, 65%); NMR d (CD_3SOCD_3) 1.28 (t, 3H), 4.30 (q, 2H), 4.40 (s, 2H), 5.81 (s, 2H), 6.88 (dd, 1H), 7.30 (m, 3H), 7.50 (d, 1H), 7.76 (s, 1H), 7.78 (d, 1H), 10.19 (brs, 1H); *M/z* (-) 439 (M^-), 437.

Example 1

Compound 2

Ethyl 4-chloroacetamido-*N*-(3,4-dichlorobenzyl)indole-2-carboxylate (0.15 g) and morpholine (2.0 ml) were dissolved in methoxyethanol (5.0 ml) and the reaction stirred for 72 hours. The reaction was then poured into water (100 ml) and the resulting solid filtered and dried *in vacuo*. The solid was dissolved in THF (2.5 ml) and methanol (2.5 ml), and to this was added NaOH (3M, 2.0 ml). The reaction was stirred for 16 hours, then concentrated. The residue was dissolved in water, and precipitated by dropwise addition of acetic acid. The resulting solid was filtered and dried *in vacuo* to give the title compound as a white solid (0.1 g, 63%, 2 steps); NMR d (CD_3SOCD_3) 2.58 (t, 4H), 3.29 (s, 2H), 3.65 (t, 4H), 5.82 (s, 2H), 6.90 (dd, 1H), 7.30 (m, 3H), 7.52 (m, 2H), 7.72 (d, 1H), 9.80 (s, 1H); *M/z* (-) 462 (M^-), 460, 418.

Example 2

The procedure described in Example 1 above was repeated using the appropriate amines. Thus were obtained the compounds described below.

Example 5

The procedure described in the Example 4 above was repeated using the appropriate acid chloride. Thus was obtained the compound described below.

Di-ester of Compound 12

64% yield; *M/z* (-) 534 (M^+), 532.

Example 6

Di-ester of Compound 14

Sarcosine ethyl ester hydrochloride (1.23 g) and potassium carbonate (1.11 g) were added to a solution of ethyl 4-chloroacetamido-*N*-(3,4-dichlorobenzyl)indole-2-carboxylate (700 mg) in acetone (25 ml), stirred and heated at 65°C overnight. The reaction was partitioned between water (50 ml) and ethyl acetate (50 ml), extracted with ethyl acetate (2 x 50 ml), and dried ($MgSO_4$). The combined organic extracts were concentrated *in vacuo*, and the residue purified by column chromatography using 30% ethyl acetate : toluene as eluent, to afford the product as a yellow solid (768 mg, 92%); NMR d (CD_3SOCD_3) 1.21 (t, 3H), 1.28 (t, 3H), 2.45 (s, 3H), 3.42 (s, 2H), 3.53 (s, 2H), 4.16 (q, 2H), 4.30 (q, 2H), 5.81 (s, 2H), 6.88 (d, 1H), 7.27 (m, 2H), 7.52 (d, 1H), 7.67 (s, 1H), 7.84 (d, 1H), 9.95 (s, 1H), *M/z*(+) 520.3 (MH^+)

Example 7

The procedure described in Example 6 above was repeated using the appropriate amine. Thus was obtained the compound described below.

Diester of Compound 13

93% yield; NMR d (CD_3SOCD_3) 1.15 (t, 3H), 1.28 (t, 3H), 3.52 (s, 3H), 3.57 (s, 3H), 3.87 (s, 2H), 4.10 (q, 2H), 4.31 (q, 2H), 5.83 (s, 2H), 6.90 (d, 1H), 7.15 - 7.44 (m, 8H), 7.53 (d, 1H), 7.67 (s, 1H), 7.83 (d, 1H); *M*/z (+) 596.5 (MH^+).

Example 8

Sodium hydride (15 mg, 60% in mineral oil) and ethyl 4-*N*-benzylglycine ethyl ester

Compound 14

60% yield; NMR d (CD_3SOCD_3) 2.46 (s, 3H), 3.38 (s, 2H), 3.42 (s, 2H), 5.88 (s, 2H), 6.92 (d, 1H), 7.20 (m, 2H), 7.31 (s, 1H), 7.50 (m, 2H), 7.82 (d, 1H); *M*/z (-) 462.2 (*M*-H$^+$).

Compound 15

15% yield; NMR d (CD_3SOCD_3) 3.21 (s, 2H), 3.31 (s, 3H), 3.40 (s, 2H), 3.69 (s, 2H), 5.83 (s, 2H), 6.90 (d, 2H), 6.98 (d, 2H), 7.15 (m, 6H), 7.27 (t, 1H), 7.39 (s, 1H), 7.53 (m, 2H); *M*/z (-) 554.3 (*M*-H$^+$).

Compound 13

25% yield; NMR d (CD_3SOCD_3) 3.44 (s, 2H), 3.46 (s, 2H), 3.85 (s, 2H), 5.91 (s, 2H), 6.87 (m, 1H), 7.13 - 7.36 (m, 6H), 7.40 (m, 2H), 7.53 (m, 2H), 7.78 (d, 1H), *M*/z (-) 538.2 (*M*-H$^+$), 253.2.

Example 11

***N*-Benzyl-4-(2-(pyrid-2-yl)thiophene-4-sulphonyl))aminoindole-2-carboxylic acid (Compound 1)**

To a solution of ethyl *N*-benzyl-4-aminoindole-2-carboxylate (140 mg) and pyridine (0.08 ml) in dichloromethane (10 ml) at 20°C was added 2-(pyrid-2-yl)thiophene-4-sulphonyl chloride (140 mg) and the reaction stirred for 2 hours. The mixture was washed with HCl (2M, 10 ml), the organic layer was concentrated *in vacuo* and the residue purified by chromatography on silica using ethyl acetate as eluent, to give a yellow solid which was dissolved in ethanol (50 ml) at 60°C and treated with NaOH (2M, 4.0 ml) with stirring for 2 hours. The solvent was evaporated *in vacuo*, the residue dissolved in water (50 ml) and filtered. The clear yellow filtrate was acidified with 2N HCl and extracted with dichloromethane / methanol (9:1, 100 ml). The organic layer was dried ($MgSO_4$) and evaporated to give a pale brown solid, which was triturated with ether to give the product as an off white powder (150 mg, 63%, 2 steps); NMR d (CD_3SOCD_3) 5.87 (s, 2H), 6.9 - 7.1 (m, 9H), 7.30 (dd, 2H), 7.43 (d, 1H), 7.63 (d, 1H), 7.81 (dd, 1H), 7.96 (d, 1H), 8.50 (d, 1H); *M*/z

(5 mg) in dichloromethane. The reaction was stirred for 16 hours at room temperature under an atmosphere of nitrogen. The reaction was washed with hydrochloric acid (2M, 70 ml), saturated aqueous sodium hydrogencarbonate solution, water and saturated sodium chloride solution. Combined organic extracts were dried ($MgSO_4$), concentrated *in vacuo* and the residue purified by column chromatography using 60% ethyl acetate : *iso*-hexane as eluent to give the product as a colourless gum (132 mg, 74%); NMR d (CD_3SOCD_3) 2.94 (s, 3H), 3.12 (s, 3H), 3.81 (s, 3H), 5.82 (s, 2H), 6.91 (m, 2H), 7.21 (s, 1H), 7.27 - 7.36 (m, 2H), 7.46 (d, 1H), 7.52 (d, 1H); *M*/z (+) 421 (MH^+).

Example 14

***N*-(3,4-Dichlorobenzyl)-4-(dimethylcarbamate)indole-2-carboxylic acid (Compound 10)**

Desesterifiation of the compound of Example 13 using the method described in Example 9 above yielded Compound 10.

93% yield; NMR d (CD_3SOCD_3) 2.94 (s, 3H), 3.11 (s, 3H), 5.91 (s, 2H), 6.82 (d, 1H), 6.94 - 7.03 (m, 2H), 7.18 (t, 1H), 7.29 - 7.39 (m, 2H), 7.50 (d, 1H); *M*/z (-) 405 (M-H^+).

Example 15

Biological Assays for hMCP-1 Antagonists

a) hMCP-1 Receptor-binding assay

i) Cloning and expression of hMCP-1 receptor

The MCP-1 receptor B (CCR2B) cDNA was cloned by PCR from THP-1 cell RNA using suitable oligonucleotide primers based on the published MCP-1 receptor sequences (Charo *et al.*, 1994, *Proc. Natl. Acad. Sci. USA*, **91**, 2752). The resulting PCR products were cloned into vector PCR-II™ (InVitrogen, San Diego, CA.). Error free CCR2B cDNA was subcloned as a Hind III-Not I fragment into the eukaryotic expression vector pCDNA3 (InVitrogen) to generate pCDNA3/CC-CKR2A and pCDNA3/CCR2B respectively.

Linearised pCDNA3/CCR2B DNA was transfected into CHO-K1 cells by calcium phosphate precipitation (Wigler *et al.*, 1979, *Cell*, **16**, 777). Transfected cells were selected by the addition of Geneticin Sulphate (G418, Gibco BRL) at 1mg/ml, 24 hours after the cells had

CHO-CCR2B was identified as the highest MCP-1 receptor B expressor.

Compounds tested of the present invention had IC_{50} values of 50μM or less in the hMCP-1 receptor binding assay described herein. For example Compound 2 in Table 1 showed IC_{50} of 1.17μM in hMCP-1.

b) MCP-1 mediated calcium flux in THP-1 cells

The human monocytic cell line THP-1 was grown in a synthetic cell culture medium RPMI 1640 supplemented with 10 % foetal calf serum, 2 mM glutamine and Penicillin-Streptomycin (at 50 μg streptomycin/ml, Gibco BRL). THP-1 cells were washed in HBSS (lacking Ca^{2+} and Mg^{2+}) + 1 mg/ml BSA and resuspended in the same buffer at a density of 3 x 10^6 cells/ml. The cells were then loaded with 1 mM FURA-2/AM for 30 min at 37°C, washed twice in HBSS, and resuspended at 1x10^6 cells/ml. THP-1 cell suspension (0.9 ml) was added to a 5 ml disposable cuvette containing a magnetic stirrer bar and 2.1 ml of prewarmed (37°C) HBSS containing 1 mg/ml BSA, 1 mM $MgCl_2$ and 2 mM $CaCl_2$. The cuvette was placed in a fluorescence spectrophotometer (Perkin Elmer, Norwalk, CT) and preincubated for 4 min at 37°C with stirring. Fluorescence was recorded over 70 sec and cells were stimulated by addition of hMCP-1 to the cuvette after 10 sec. $[Ca^{2+}]i$ was measured by excitation at 340 nm and 380 nm alternately and subsequent measurement of the intensity of the fluorescence emission at 510 nm. The ratio of the intensities of the emitted fluorescent light following excitation at 340 nm and 380 nm, (R), was calculated and displayed to give and estimate of cytoplasmic $[Ca^{2+}]$ according to the equation:-

$$[Ca^{2+}]i = K_d \frac{(R-Rmin)}{(Rmax-R)} (Sf2/Sb2)$$

where the K_d for FURA-2 Ca^{2+} complex at 37°C was taken to be 224 nm. R_{max} is the maximal fluorescence ratio determined after addition of 10 mM Ionomycin, R_{min} is the minimal ratio determined by the subsequent addition of a Ca^{2+} free solution containing 5 mM EGTA, and Sf2/Sb2 is the ratio of fluorescence values at 380 nm excitation determined at R_{min} and R_{max}, respectively.

Stimulation of THP-1 cells with hMCP-1 induced a rapid, transient rise in $[Ca^{2+}]i$ in a specific and dose dependent manner. Dose response curves indicated an approximate EC_{50} of 2 nm. Test compounds dissolved in DMSO (10μl) were assayed for inhibition of calcium

agonism by addition in place of hMCP-1

hMCP-1 induced concentration dependent cell migration with a characteristic biphasic response, maximal 0.5-1.0 nm.

In an alternative form of the above assay, fluorescently tagged cells can be used in order to assist in end point detection. In this case, the THP-1 cells used are fluorescently tagged by incubation in the presence of 5mM Calcein AM (Glycine, N,N'-[[3',6'-bis(acetyloxy)-3-oxospiro[isobenzofuran-1(3H),9'-[9H]xanthene]-2',7'-diyl]bis(methylene)]bis[N-[2-[(acetyloxy)methoxy]-2-oxoethyl]]-bis[(acetyloxy)methyl] ester; Molecular Probes) for 45 minutes in the dark. Cells are harvested by centrifugation and resuspended in HBSS (without Phenol Red) with Ca2+, Mg2+ and 0.1% BSA. 50ml (2x105 cells) of the cell suspension are placed on the filter above each well and, as above, the unit is incubated at 37°C for 2 hours under 5% CO2. At the end of the incubation, cells are washed off the upper face of the filter with phosphate buffered saline, the filter removed from the plate and the number of cells attracted to either the underside of the filter or the lower well estimated by reading fluorescence at 485nm excitation, 538nm emission wavelengths (fmax, Molecular Devices). The data was input into a spreadsheet, corrected for any random migration in the absence of chemoattractant and the average fluorescence values, standard error of the mean, percentage inhibition and IC50 of compounds under test and significance tests can be calculated .

Compound No. 13 in Table I showed 94% inhibition at 20μm.

No physiologically unacceptable toxicity was observed at the effective dose for compounds tested of the present invention.

Example 16

Pharmaceutical Compositions

The following Example illustrates, but is not intended to limit, pharmaceutical dosage forms of the invention as defined herein (the active ingredient being termed "Compound X"), for therapeutic or prophylactic use in humans:

(e)

Injection I	(50 mg/ml)
Compound X	5.0% w/v
1M Sodium hydroxide solution	15.0% v/v
0.1M Hydrochloric acid	to adjust pH to 7.6
Polyethylene glycol 400	4.5% w/v
Water for injection	to 100%

(f)

Injection II	(10 mg/ml)
Compound X	1.0% w/v
Sodium phosphate BP	3.6% w/v
0.1M Sodium hydroxide solution	15.0% v/v
Water for injection	to 100%

(g)

Injection III	(1mg/ml, buffered to pH6)
Compound X	0.1% w/v
Sodium phosphate BP	2.26% w/v
Citric acid	0.38% w/v
Polyethylene glycol 400	3.5% w/v
Water for injection	to 100%

(h)

Aerosol I	mg/ml
Compound X	10.0
Sorbitan trioleate	13.5
Trichlorofluoromethane	910.0

Note:

Compound X in the above formulation may comprise a compound illustrated in Examples. The above formulations may be obtained by conventional procedures well known in the pharmaceutical art. The tablets (a)-(c) may be enteric coated by conventional means, for example to provide a coating of cellulose acetate phthalate. The aerosol formulations (h)-(k) may be used in conjunction with standard, metered dose aerosol dispensers, and the suspending agents sorbitan trioleate and soya lecithin may be replaced by an alternative suspending agent such as sorbitan monooleate, sorbitan sesquioleate, polysorbate 80, polyglycerol oleate or oleic acid.

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(54) Title: POTENTIATION OF NMDA ANTAGONISTS

(57) Abstract

The present invention is directed to a method for potentiating the therapeutic effects of selected NMDA antagonists.

POTENTIATION OF NMDA ANTAGONISTS

The present invention is directed to a method for potentiating the therapeutic effects of a group of excitatory amino acid antagonists. Another aspect of the invention is directed to new pharmaceutical compositions useful for the treatment of conditions associated with excessive excitatory amino acid activity.

In accordance with the present invention, it has been discovered that probenecid will potentiate the activity of the following excitatory amino acid antagonists:

a) in the compounds of Formula Ia, X is represented by a linear C_{1-4} alkylene, or S; m is an integer from 1-4; Z is represented by H, C_{1-4} alkyl, phenyl, substituted phenyl, or an alkylphenyl substituent in which the phenyl ring may be optionally substituted; R is represented by hydrogen, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, CF_3, OCF_3, OH, NO_2, or CN; R_1 and R_2 are each independently represented by -OH, $-OR_3$, $-NR_4R_5$, $-OCH_2OR_3$, or $-O-(CH_2)_n-NR_6R_7$, in which n is an integer from 1-4; R_3 is represented by C_{1-4} alkyl, phenyl, substituted phenyl or an alkylphenyl substituent in which the phenyl ring may be optionally substituted; R_4 and R_5 are each independently represented by hydrogen or a C_{1-4} alkyl; R_6 and R_7 are each independently represented by hydrogen or a C_{1-4} alkyl, or R_6 and R_7 together with the adjacent nitrogen atom form a piperidino, morpholino, or pyrrolidino group with the proviso that if X is a C_{1-4} alkylene, then m is O;

b) in the compounds of Formula Ib, R, Z, and R_2 are as above, B is represented by hydrogen, C_1-C_4 alkyl, optionally substituted alkylphenyl, or $-CH_2-COR_2$; Y is SO_2 or CO; and A is represented by phenyl, substituted phenyl, or C(O)D in which D is defined as R_2 above;

c) in the compounds of Formula Ic, E is represented by hydrogen, C_{1-4} alkyl, or $-CF_3$; A is represented by a methylene or a trimethylene bridging group; and E_1 is represented by hydrogen, C_{1-4} alkyl, cycloalkyl, trialkylamino, alkylphenyl, phenyl, or substituted phenyl;

d) in the compounds of Formula Id, B and E_1 are as above,

CF_3, OCF_3, OH, and CN. These substituents may be the same or different and may be located at any of the ortho, meta, or para positions;

e) the term "alkylphenyl substituent" refers to the following structure $-(CH_2)_m-C_6H_5$, in which m is an integer from 1-3. This phenyl ring may be substituted in the manner described immediately above;

h) the term "pharmaceutically acceptable addition salt" refers to either a pharmaceutically acceptable acid addition salt or a pharmaceutically acceptable basic addition salt;

i) the term "halogen" refers to a fluorine, bromine or chlorine atom;

j) the term "trialkylamine" refers to $-(CH_2)_n-N(Alk)-Alk_1$ in which n is represented by an integer from 2-4 and Alk and Alk_1 are each independently represented by a C_1-C_4 alkyl; and

k) the term "cyclohexylmethyl" refers to $-CH_2-C_6H_{12}$.

The expression "pharmaceutically acceptable basic addition salts" is intended to apply to any non-toxic organic or inorganic basic addition salts of the compounds represented by Formulae Ia-d or any of its intermediates. Illustrative bases which form suitable salts include alkali metal or alkaline-earth metal hydroxides such as sodium, potassium, calcium, magnesium, or barium hydroxides; ammonia, and aliphatic, alicyclic, or aromatic organic [illegible]amine,

specific optical isomer or a mixture of optical isomers (unless it is expressly excluded). The specific optical isomers can be separated and recovered by techniques known in the art such as chromatography on chiral stationary phases or resolution via chiral salt formation and subsequent separation by selective crystallization. Alternatively utilization of a specific optical isomer as the starting material will produce the corresponding isomer as the final product.

As is indicated by the E_2 substituent in the compounds of formula Id, the piperidine ring may be further substituted at positions 4, 5, or 6. E_2 may optionally represent up to 2 non-hydrogen substituents. Only one non-hydrogen substituent should be located at any one position on the piperidine ring. If two non-hydrogen substituents are present, they may be the same or different. When E_2 is a non-hydrogen substituent, then this substituent may be either syn or anti relative to the phosphono substituent.

All of the compounds of Formula Id contain at least two (2) asymetric centers and thus will exist as diasteriosmers. Any reference to these compounds as well as their intermediates should be construed as encompassing a racemic mixture, a specific optical isomer or a pair of enantiomers. The specific optical isomers can be synthesized as shown herein or can be recovered by techniques known in the art such as chromatography on chiral stationary phases, or resolution via chiral salt formation and subsequent separation by selective crystallization. HPLC ion exchange chromatography may be utilized to separate only the diastereomers.

[illegible] compounds

acid antagonists, and methods for preparing pharmaceutical formulations from them. Preferred compounds are those in which R is represented by a 4,6-dihalo substituent.

The compounds of Formula Ib are the subject of United States Patent Application No. 07/608,457, filed November 2, 1990, which is hereby incorporated by reference. This application describes methods for their synthesis, their use as excitatory amino acid antagonists and pharmaceutical formulations containing them. Preferred compounds are those in which R is a 4,6-dihalo substituent, B is alkyl, Z is hydrogen and A is phenyl. The most preferred compound is 3-[(phenacyl)methylamino]-2-carboxy-4,6-dichloroindole.

The compounds of Formula Ic are known in the art and are described in European Patent Application No. 0 418 863 as well as its US counterpart, Patent Application No. 553,431 filed July 20, 1990, now allowed, which is hereby incorporated by reference. These applications describe methods for their synthesis, their use as excitatory amino acid antagonists and pharmaceutical formulations containing them. Preferred compounds include those in which A is methylene and E and E_1 are hydrogen. The most preferred compound is R-4-Oxo-5-phosphonohomovaline.

The compounds of Formula Id are the subject of United States Patent Application No. 525, 290 filed May 17, 1990 which is hereby incorporated by reference. This application discloses methods for their synthesis, their use as excitatory amino acid antagonists and pharmaceutical formulations containing them. Preferred compounds include those in which the stereochemistry is 2R,3S and in which B_5 is hydrogen or 4-alkyl. Preferred compounds include 3-

cord, and neonatal anoxic trauma. The compounds should be administered to the patient within 24 hours of the onset of the hypoxic, ischemic, or hypoglycemic condition in order for the compounds to effectively minimize the CNS damage which the patient will experience.

The compounds exhibit anti-convulsant properties and are useful in the treatment of epilepsy. They are useful in the treatment of grand mal seizures, petit mal seizures, psychomotor seizures, and autonomic seizures. The compounds are also useful in the treatment of neurodegenerative diseases such as Huntington's disease, Alzheimer's disease, senile dementia, glutaric acidaemia type I, multi-infarct dementia, and neuronal damage associated with uncontrolled seizures. The administration of these compounds to a patient experiencing such a condition will serve to either prevent the patient from experiencing further neurodegeneration or it will decrease the rate at which the neurodegeneration occurs. As is apparent to those skilled in the art, the compounds will not correct any CNS damage that has already occurred as the result of either disease or a lack of oxygen or sugar. As used in this application, the term "treat" refers to the ability of the compounds to prevent further damage or delay the rate at which any further damage occurs. The compounds may also be utilized as anxiolytic agents and as analgesics. The therapeutic activity of these compounds is described in more detail in the United States patents and patent applications which were incorporated by reference above.

The compounds may be administered concurrently with probenecid in order to treat any [illegible] diseases or [illegible] of probenecid that

Table I

COMPOUNDS	DOSAGE RANGE (Mg/Kg-day)
Ia	0.1 - 50
Ib	0.1 - 50
Ic	1 - 500
Id	0.1 - 500

With the concurrent administration of probenecid, this dosage range may be adjusted lower by a factor of from 2- to 10-fold. Alternatively, the compounds may be administered at the same dosage range in order to obtain an enhanced effect due to the higher therapeutic concentrations obtained. The dosage frequency of the compounds can vary widely depending upon the condition or disease being treated. Repetitive daily administration may be desirable and will vary according to the conditions outlined above. For certain conditions such as stroke, it may be desirable to maintain a continuous IV infusion.

Probenecid is currently available commercially as tablets. The sodium salt of probenecid is readily water soluble and injectable dosage forms can be prepared from this salt using techniques well known to those skilled in the art.

The compounds may be administered by a variety of routes. They are effective if administered orally. The compounds may also be administered parenterally (i.e.

carrier and administered as either a solution or a suspension. Illustrative of suitable pharmaceutical carriers are water, saline, dextrose solutions, fructose solutions, ethanol, or oils of animal, vegetative, or synthetic origin. The pharmaceutical carrier may also contain preservatives, buffers, etc., as are known in the art. When the medicaments are being administered intrathecally, they may also be dissolved in cerebrospinal fluid as is known in the art.

As used in this application:

a) the term "patient" refers to warm blooded animals such as, for example, guinea pigs, mice, rats, cats, rabbits, dogs, monkeys, chimpanzees, and humans;

b) the term "treat" refers to the ability of the compounds to either relieve, alleviate, or slow the progression of the patient's disease;

c) the term "neurodegeneration" refers to a progressive death and disappearance of a population of nerve cells occurring in a manner characteristic of a particular disease state and leading to brain damage;

d) the phrase "concurrent administration" refers to administering the probenicid at an appropriate time so that it will potentiate the antagonistic effects of the compounds of Formula I. This may means simultaneous administration or administration at appropriate but different times. Establishing such a proper dosing schedule will be readily apparent to one skilled in the art.

B) The protocol described above was repeated with minor variations with the compound 3-[(carbethoxymethyl)thio]-2-carbethoxy-4,6-dichloroindole. The test was conducted in the following manner.

Groups of DBA/2J audiogenic mice were administered i.p. 6 doses ranging from 25 to 400 mg/kg of 3-[(carboxymethyl)thio]-2-carboxy-4,6-dichloroindole (hereinafter compound). Five minutes after administration, they were placed individually in glass jars and exposed to a sound stimulus of 110 decibels for 30 seconds. Each mouse was observed during the sound exposure for signs of seizure activity. A graph was prepared based upon the dose administered and the percentage of animals protected at that dose. An ED_{50} was calculated from the graph. The test was also performed in separate mice with the only modification being the addition of 100mg/kg or 200mg/kg of probenecid IP. The following ED_{50}'s were obtained.

TREATMENT	ED_{50} (mg/kg)
Compound	149
Compound + 100 mg/kg probenecid	45.2
Compound + 200 mg/kg	11.0

EXAMPLE II

The intracerebroventricular administration of quinolinic [illegible] clonic seizures in mice. If a compound can

WHAT IS CLAIMED IS:

1. A pharmaceutical composition suitable for antagonizing the effects of excitatory amino acids upon the NMDA receptor complex comprising an effective amount of probenecid and an antagonistic amount of a compound of the formulae:

by H, C_{1-4} alkyl, phenyl, substituted phenyl, or an alkylphenyl substituent in which the phenyl ring may be optionally substituted; R is represented by hydrogen, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, CF_3, OCF_3, OH, NO_2, or CN; R_1 and R_2 are each independently represented by $-OH$, $-OR_3$, $-NR_4R_5$, $-OCH_2OR_3$, or $-O-(CH_2)_n-NR_6R_7$, in which n is an integer from 1-4; R_3 is represented by C_{1-4} alkyl, phenyl, substituted phenyl or an alkylphenyl substituent in which the phenyl ring may be optionally substituted; R_4 and R_5 are each independently represented by hydrogen or a C_{1-4} alkyl; R_6 and R_7 are each independently represented by hydrogen or a C_{1-4} alkyl, or R_6 and R_7 together with the adjacent nitrogen atom form a piperidino, morpholino, or pyrrolidino group; with the proviso that if X is a C_{1-4} alkylene, then m is O;

b) in the compounds of Formula Ib, R, Z, and R_2 are as above, B is represented by hydrogen, C_1-C_4 alkyl, optionally substituted alkylphenyl, or $-CH_2-COR_2$; Y is SO_2 or CO; A is represented by phenyl, substituted phenyl, or C(O)D in which D is defined as R_2 above;

c) in the compounds of Formula Ic, E is represented by hydrogen, C_{1-4} alkyl, or $-CF_3$; A is represented by a methylene or a trimethylene bridging group; and E_1 is represented by hydrogen, C_{1-4} alkyl, cycloalkyl, trialkylamino, alkylphenyl, phenyl, or substituted phenyl;

d) in the compounds of Formula Id E and E_1 are as above, E_2 is represented by hydrogen, C_1-C_4 alkyl, phenyl, alkylphenyl, or cyclohexylmethyl;
E_5 is represented by hydrogen, linear C_1-C_4 alkyl, or alkylphenyl; or a pharmaceutically acceptable salt of any of the compounds of Formulae Ia-d, in admixture with a

INTERNATIONAL SEARCH REPORT

International application No.
PC[redacted]S 92/10354

A. CLASSIFICATION OF SUBJECT MATTER

IPC5: A61K 31/19, A61K 31/405, A61K 31/445, A61K 31/66
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC5: A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

CA, MEDLINE, EMBASE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	GB, A, 2100127 (SANTEN PHARMACEUTICAL CO LTD), 22 December 1982 (22.12.82), page 1, line 7 - line 17	1-6
	--	
Y	US, A, 4960786 (F G SALITURO ET AL), 2 October 1990 (02.10.90)	1-6
	--	
Y	US, A, 5051442 (F G SALITURO ET AL), 24 Sept 1991 (24.09.91)	1-6
	--	

[X] Further documents are listed in the continuation of Box C. [X] See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"E" erlier document but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

[illegible] March [illegible]

Name and mailing address of the ISA:
European Patent Office, P.B. 5818 Patentlaan 2
NL-2280 HV Rijswijk

Authorized officer

CAROLINA GOMEZ LAGERLÖF

SA 7478

INTERNATIONAL SEARCH REPORT

29/01/93

International application No. PCT/US 92/10354

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
GB-A- 2100127	22/12/82	BE-A- 890398	18/01/82
		DE-T- 3152363	21/11/85
		FR-A,B- 2490490	26/03/82
		JP-C- 1406415	27/10/87
		JP-A- 57056425	05/04/82
		JP-B- 62013923	30/03/87
		SE-A- 8203054	14/05/82
		US-A- 4424228	03/01/84
		WO-A- 8200954	01/04/82
US-A- 4960786	02/10/90	AU-B- 625998	23/07/92
		AU-A- 5378090	25/10/90
		EP-A- 0394905	31/10/90
		JP-A- 2295924	06/12/90
US-A- 5051442	24/09/91	AU-A- 7650091	11/11/91
		WO-A- 9116307	31/10/91
WO-A1- 9215565	17/09/92	AU-A- 1532092	06/10/92

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
PHM 70471/WO		
International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/GB00/00260	31/01/2000	05/02/1999
International Patent Classification (IPC) or national classification and IPC		
C07D209/42		
Applicant		
ASTRAZENECA AB et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 5 sheets, including this cover sheet.

   [ ] This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of sheets.

3. This report contains indications relating to the following items:

   I [X] Basis of the report
   II [ ] Priority
   III [ ] Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   IV [ ] Lack of unity of invention
   V [X] Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   VI [ ] Certain documents cited
   VII [X] Certain defects in the international application
   VIII [X] Certain observations on the international application

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/GB00/00260

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:
Description, pages:

1-39 as originally filed

Claims, No.:

1-10 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).

☐ the language of publication of the international application (under Rule 48.3(b)).

☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

[ ] contained in the international application in written form.

☐ filed together with the international application in computer readable form.

☐ furnished subsequently to this Authority in written form.

[ ] furnished subsequently to this Authority in computer readable form

[ ] The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

[ ] The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

the description, pages

(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-10
	No:	Claims	
Inventive step (IS)	Yes:	Claims	1-10
	No:	Claims	
Industrial applicability (IA)	Yes:	Claims	1-9
	No:	Claims	10 see below

2. Citations and explanations
see separate sheet

VII. Certain defects in the international application

The following defects in the form or contents of the international application have been noted:
see separate sheet

VIII. Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:
see separate sheet

V. CITATIONS AND EXPLANATIONS

The following documents are mentioned in this report.

US-A-5,288,743	(A)
WO-A-99 33800	(B)
WO-A-99 07678	(C)
WO-A-99 07351	(D)

The novel feature of the indole derivative of claim 1 is the R4 group, representing an acylamino, sulfonylamino or aminocarbonyloxy group, present at the 4-position of the ring. The dependent claims 2-7, as well as claim 8 drawn to a process for the preparation of compounds of claim 1, and claims 9 and 10 drawn to pharmaceutical compositions containing compounds of claim 1 and compounds of claim1 for use in the preparation of medicaments are novel by consequence. Claims 1 to 10 therefore meet the Novelty requirements of Article 33(2) PCT.

Document (A) represents the closest prior art. This document describes some 1-benzyl-2-carboxyalkyl-5-(heterocyclylmethoxy)- indoles and their use for the inhibition of leukotriene synthesis. the compounds of document (A) are useful for the treatment of inflammation (see column 2, lines 15-20). The presently claimed compounds also have anti-inflammation activity. and differ from the compounds of document (A) through the absence of an alkyl group linking the carboxy group to the 2-position, and through the presence of the R4 group as defined above at the 4-position of the indole ring. Hence the presently claimed compounds are not structurally close to the compounds of document (A), and it would not have been obvious for the skilled man to prepare them in order to make available further anti-inflammation compounds. Inventive step (Article 33(3) PCT) is recognise because the problem of providing further anti-inflammation compounds has been solved in a non obvious manner.

For the assessment of the present claim 10 on the question whether it is industrially [illegible] exist in the PCT Contracting States. The patentability can

for first use in medical treatment and the use of such a compound for the manufacture of a medicament for a new medical treatment.

At present no priority document is available. The examination has been carried out assuming that the priority date is validly claimed. If during the subsequent procedure (e.g. EPO examination) the priority date is found to be invalid for some or all of the presently claimed subject matter, the intermediate documents (B)-(D) may be taken into consideration for the evaluation of Novelty and inventive step.

VII CERTAIN DEFECTS IN THE INTERNATIONAL APPLICATION.

Contrary to the requirements of Rule 5.1(a)(ii) PCT, the relevant background art disclosed in the documents (A) and (B) is not mentioned in the description, nor are these documents identified therein.

VIII. CERTAIN OBSERVATIONS ON THE INTERNATIONAL APPLICATION.

Claim 1 contains several non-limiting definitions such as "optionally substituted aryl" and "optionally substituted heteroaryl", etc. which embrace substitution by any known organic group without limitation on size or number of reactive groups which can be present. The term "heteroaryl" itself embraces any known aromatic heterocyclic group. It is known in pharmaceutical chemistry that small structural changes to heterocyclic rings can lead to considerable changes in a pharmacological activity, or to compounds with a completely different activity. The skilled man would therefore not be able to predict if all compounds falling within the said definition "heteroaryl" would actually solve the problem underlying the present application (i.e. the provision of MCP-1 inhibitors). Also, since the term "functional group" appears to embrace any reactive group and is not limited to the groups suggested on page 4, lines 10-14, it is not clear if the presence of any "functional group" at R4-R7 would give rise to a compound which binds to a MCP-1 receptor, because some reactive groups would be expected to react at competing binding sites.